JC17 Rec'd PCT/PTO 71 1 MAY 2001 U.S. DEPARTMENT OF COMMERCE PATENT AND TRADEMARK OFFICE ATTORNEY'S DOCKET NUMBER 06501-080001 TRANSMITTAL LETTER TO THE UNITED STATES DESIGNATED/ELECTED OFFICE (DO/EO/US) U.S. APPLICATION NO. (If Known, see 37 CFR **CONCERNING A FILING UNDER 35 U.S.C. 371** INTERNATIONAL FILING DATE PRIORITY DATE CLAIP INTERNATIONAL APPLICATION NO. PCT/JP99/04520 23 August 1999 13 November 1998 TITLE OF INVENTION HYBRIDIZATION PROBE APPLICANT(S) FOR DO/EO/US Tomoyasu Sugiyama, Toshio Ota, Shizuko Ishii and Ai Wakamatsu Applicant herewith submits to the United States Designated/Elected Office (DO/EO/US) the following items and other information: This is a FIRST submission of items concerning a filing under 35 U.S.C. 371. This is a **SECOND** or **SUBSEQUENT** submission of items concerning a filing under 35 U.S.C. 371. his is an express request to promptly begin national examination procedures (35 U.S.C. 371(f)). The US has been elected by the expiration of 19 months from the priority date (PCT Article 31). A copy of the International Application as filed (35 U.S.C. 371(c)(2)) is attached hereto (required only if not communicated by the International Bureau). A has been communicated by the International Bureau. is not required, as the application was filed in the United States Receiving Office (RO/US). 6. An English language translation of the International Application as filed (35 U.S.C. 371(c)(2)). Amendments to the claims of the International Application under PCT Article 19 (35 U.S.C. 371(c)(3)) are attached hereto (required only if not communicated by the International Bureau). have been communicated by the International Bureau. have not been made; however, the time limit for making such amendments has NOT expired. d. M have not been made and will not be made. 8. An English language translation of amendments to the claims under PCT Article 19 (35 U.S.C. 371(c)(3)). 9. An oath or declaration of the inventor(s) (35 U.S.C. 371(c)(4)). 10. An English language translation of the annexes to the International Preliminary Examination Report under PCT Article 36 (35 U.S.C. 371(c)(5)). Items 11 to 16 below concern other documents or information included: 11. An Information Disclosure Statement under 37 CFR 1.97 and 1.98. 12. An assignment document for recording. A separate cover sheet in compliance with 37 CFR 3.28 and 3.31 is included. 13. A FIRST preliminary amendment.

CERTIFICATE OF MAILING BY EXPRESS MAIL

Express Mail Label No EL624321289US

I hereby certify under 37 CFR §1 10 that this correspondence is being deposited with the United States Postal Service as Express Mail Post Office to Addressee with sufficient postage on the date indicated below and is addressed to the Commissioner for Patents, Washington, D C 20231

May 11, 2001

Date of Deposit

A SECOND or SUBSEQUENT preliminary amendment.

15. A change of power of attorney and/or address letter.

14. A substitute specification.

16. Other items or information:

Typed Name of

U.S. APPLICATION OF (IF KI	877591	PCT/JP99/04520	ATION NO.	06501-080001	THOMBER	
17. 🖂 The following fees are submitted:				CALCULATIONS ONLY	CALCULATIONS PTO USE	
Basic National Fee ( 37 CFR 1.492(a)(1)- (5) ):				UNLT		
Neither international preliminary examination fee (37 CFR 1.482)						
nor international search fee (37 CFR 1.445(a)(2)) paid to USPTO and International Search Report not prepared by the EPO or JPO <b>\$1000</b>						
International preliminary						
USPTO but International Search Report prepared by the EPO or JPO \$860						
International preliminary examination fee (37 CFR 1.482) not paid to USPTO but international search fee (37 CFR 1.445(a)(2)) paid to USPTO						
International preliminary examination fee paid to USPTO (37 CFR 1.482) but all claims did not satisfy provisions of PCT Article 33(1)-(4)						
International preliminary and all claims satisfied p	examination fee pa provisions of PCT A	aid to USPTO (37 CFF	R 1.482) <b>\$100</b>			
ENTER APPROPRIATE BASIC FEE AMOUNT =				\$860.00		
Surcharge of \$130 for furnishing the oath or declaration later than   20 30 30 months from the earliest claimed priority date (37 CFR 1.492(e)).				\$0.00		
Claims	Number Filed	Number Extra	Rate			
Total Claims	16 - 20 =	0	x \$18	\$0.00		
Independent Claims	4 - 3 =	11	x \$80	\$80.00		
MULTIPLE DEPENDEN			+ \$270	\$270.00		
TOTAL OF ABOVE CALCULATIONS =				\$1,210.00		
Applicant claims small entity status. See 37 CFR 1.27. The fees indicated above are				\$605.00		
reduced by 1/2.				\$605.00 \$605.00		
Processing fee of \$130 for furnishing the English Translation later than 20 30				φυσσ.σσ		
months from the earliest claimed priority date (37 CFR 1.492(f))				\$0.00		
TOTAL NATIONAL FEE =				\$605.00		
Fee for recording the enclosed assignment (37 CFR 1.21(h)). The assignment must be				<b>60.00</b>		
accompanied by an appropriate cover sheet (37 CFR 3.28, 3.31).\$40.00 per property +				\$0.00 \$605.00		
TOTAL FEES ENGLOSED -				Amount to be	\$	
				refunded:		
f				Charged:	\$	
b. Please charge r	ny Deposit Account	to cover the above fee t No. 06-1050 in the a	es is enclosed. mount of \$0.00 to cover			
copy of this sheet is enclosed.  c.   The Commissioner is hereby authorized to charge any additional fees which may be required, or credit any overpayment to Deposit Account No. 06-1050. A duplicate copy of this sheet is enclosed.						
, ,	•				•	
NOTE: Where an appr (37 CFR 1.137(	opriate time limit ( a) or (b) must be f	under 37 CFR 1.494 ( iled and granted to r	or 1.495 has not been mestore the application t	o pending status.	evive	
SEND ALL CORRESPONDE	NCE TO:			(		
Janis K. Fraser, Ph.D., J.D.						
FISH & RICHARDSON P.C.  225 Franklin Street				\	5t D 10	
Boston, MA 02110-2804 NAME				Janis K. Fraser,	Pn.D., J.D.	
(617) 542-5070 phone				34,81	Q	
(617) 542-8906 facsimile REGISTRATION NUMBER						

2/PRTS

## DESCRIPTION

## HYBRIDIZATION PROBE

## 5 Technical Field

The present invention relates to a labeled DNA and a method for detecting genes with the labeled DNA.

## Background Art

Labeled DNAs have been utilized in a wide variety of analytical methods based on the specific interaction between DNAs having complementary nucleotide sequences. The following DNA-labeling methods are known:

- (1) a method in which a radioisotope-labeled phosphate group is introduced into the 5' end of synthetic DNA with T4 polynucleotide kinase;
- (2) a method in which biotin or digoxigenin is directly added to the end of oligo DNA by a chemical method, thereby labeling the DNA; or
- (3) a method in which a labeled nucleotide is added to an oligo DNA with terminal transferase or the like after the DNA is synthesized.

However, it is difficult to prepare probes having high specific activity by the method (1) or (2) of these methods. Only one label molecule per DNA molecule can be introduced by the methods, because the label is linked to only an end of the DNA molecule according to the methods. Therefore, high specific activity cannot be expected based on the methods.

On the other hand, multiple nucleotides are connected together in the method of (3), which is also referred to as 3'-tailing label method. A number of labeled nucleotides or nucleotide derivatives (hereafter, the term "nucleotide compounds", which means both nucleotides and nucleotide derivatives, is used in some cases) can be introduced into a DNA and can achieve high specific activity. It is sometimes difficult to introduce multiple nucleotide derivatives by this method because of steric hindrance by the nucleotide derivatives to be introduced. However, the problem of steric

20

25

30

35